

CIRCULAR TEMPLATES AND METHODS

ABSTRACT OF THE DISCLOSURE

The enzyme controlled synthesis of nucleic acids directed by a DNA template is the model of fidelity and regiospecificity for designing template-directed reactions in organic synthesis. This research has focused on the design, synthesis and evaluation of methodology for template-directed chemical ligations and oligomerizations of DNA oligonucleotides and mononucleotides. The use of circularized DNA as the template for directing the assembly of desired reaction substrates has been investigated. The effect of primers, primer length, concentrations, buffer, salts, pH, temperature, coupling reagent, and coreagents has been investigated. The difference between ligation of 3'-phosphates and 5'-phosphates has also been investigated. The present optimal conditions for the ligation and oligomerization reactions has resulted. In all cases investigated to date, the ligation of short DNA oligonucleotides on circularized DNA templates is always more efficient than the corresponding ligations on single-strand Watson-Crick DNA templates.